variety of diseases and conditions. The amendments are designed to more particularly point out and distinctly claim the subject matter that Applicant regards as the invention and not to alter the scope thereof. Claims 65, 72 and 86 are amended to correct inadvertent obvious typographical errors. None of the amendments that have been made in this response or previous responses are designed to avoid art nor to alter the scope of the claims, which scope remains as originally filed.

Attached are marked up claims pursuant to 37 C.F.R. §1.121 and copies of references to which this response refers.

DECLARATION

The Examiner indicates that the DECLARATION under 37 C.F.R. §1.132 was not received. It was filed with the Supplemental Response, mailed on September 11, 2000, and received by the PTO on September 13, 2000. A copy of the stamped received Postcard, Supplemental Response, and executed DECLARATION are attached to this response.

INFORMATION DISCLOSURE STATEMENTS

The Examiner indicates that a number of references are missing from the Information Disclosure Statement. The undersigned assures the Examiner that all cited references have been provided to the Office at least once in connection with this case or a parent thereof. In the interest of ensuring a complete record, staff members from the office of the undersigned will contact the Examiner to ascertain what references are missing from the Information Disclosure Statements filed prior to the date of the Office Action and subsequent to the Action. Arrangements for hand delivery of any missing items and references will be made.

THE REJECTION OF CLAIMS 29, 40, 43-45, 53,55, 56 AND 63 UNDER 35 U.S.C. §112, SECOND PARAGRAPH

Claims 65, 68, 69 and 71 are rejected under 35 U.S.C. § 112, second paragraph, as allegedly being indefinite for failing to particularly point out and distinctly claim the subject matter which applicant regards as the invention

because of an inadvertent typographical error in claim 65. It is respectfully submitted that the amendment of claim 65 herein to correct the typographical error obviates the grounds for this rejection.

THE REJECTION OF CLAIMS 26-29, 31, 32, 34-38, 40, 42, 44, 46, 48-54, 57 and 65-87 UNDER 35 U.S.C. § 112, FIRST PARAGRAPH

A. Claims 26-29, 31, 32, 34-38, 40, 42, 44, 46, 48-54, 57 and 65-87 are rejected under 35 U.S.C. §112, first paragraph for the reasons already of record on pages 5 and 6 of the Office Action dated 3/2/00. The Examiner states:

> the breadth of the claims is large with regard to a method of treating disorders associated with inflammatory responses associated with activation, proliferation and/or migration of immune effector cells. In addition, Applicants provide no guidance of how to treat every possible disorder associated with an inflammatory response.

Thus the Examiner urges that the methods claim treatment of a range of seemingly unrelated diseases. This rejection is respectfully traversed.

Relevant law

To satisfy the enablement requirement of 35 U.S.C § 112, first paragraph, the specification must teach one of skill in the art to make and use the invention without undue experimentation. Atlas Powder Co. v. E.I. DuPont de Nemours, 750 F.2d 1569, 224 USPQ 409 (1984). This requirement can be met by providing sufficient disclosure, either through illustrative examples or terminology, to teach one of skill in the art how to make and how to use the claimed subject matter without undue experimentation. This clause does not require "a specific example of everything *within the scope* of a broad claim." In re Anderson, 176 USPQ 331, at 333 (CCPA 1973), emphasis in original. Rather, the requirements of § 112, first paragraph "can be fulfilled by the use of illustrative examples or by broad terminology." In re Marzocci et al., 469 USPQ 367 (CCPA 1971)(emphasis added).

Further, because "it is manifestly impracticable for an applicant who discloses a generic invention to give an example of every species falling within

it, or even to name every such species, it is sufficient if the disclosure teaches those skilled in the art what the invention is and how to practice it." In re Grimme, Keil and Schmitz, 124 USPQ 449, 502 (CCPA 1960). Thus, there is no doubt that a patentee's invention may be broader than the particular embodiment shown in the specification. A patentee not only is entitled to narrow claims particularly directed to the preferred embodiment, but also to broad claims that define the invention without a reference to specific instrumentalities. Smith v. Snow, 294 U.S. 1, 11, 24 USPQ 26, 30 (1935).

Thus, there is no requirement for disclosure of every species within a genus. Applicant is entitled to claims are commensurate in scope not only with what applicant has specifically exemplified, but commensurate in scope with that which one of skill in the art could obtain by virtue of that which the applicant has disclosed.

The inquiry with respect to scope of enablement under 35 U.S.C. §112, first paragraph, is whether it would require undue experimentation to make and use the claimed invention. A considerable amount of experimentation is permissible, particularly if it is routine experimentation. The amount of experimentation that is permissible depends upon a number of factors, which include: the quantity of experimentation necessary, the amount of direction or guidance presented, the presence or absence of working examples, the nature of the invention, the state of the prior art, the relative skill of those in the art, the predictability of the art, and the breadth of the claims. Ex parte Forman, 230 USPQ 546 (Bd. Pat. App. & Int'f 1986); see also In re Wands, 8 USPQ2d 1400 (Fed. Cir. 1988).

PTO GUIDELINES

The standard for determining whether the specification meets the enablement requirement is whether it enables any person skilled in the art to make and use the claimed subject matter without undue experimentation. In re Wands, 858 F.2d 731, 737, 8 USPQ2d 1400 (Fed. Cir. 1999) (emphasis

added). In determining whether any experimentation is "undue," the above-noted factors are to be considered.

As instructed in the published PTO guidelines, it is improper to conclude that a disclosure is not enabling based on an analysis of only one of the above factors while ignoring one or more of the others. The analysis must consider all the evidence related to each of the factors, and any conclusion of non-enablement must be based on the evidence as a whole. Id. 8 USPQ2d at 1404 & 1407.

The starting point in an evaluation of whether the enablement requirement is satisfied is an analysis of each claim to determine its scope. As set forth in the guidelines, all questions of enablement are evaluated against **the claimed** subject matter. The focus of the inquiry is whether everything within the scope of the claim is enabled. With respect scope of enablement, the only relevant concern should be whether the scope of enablement provided to one skilled in the art by the disclosure is commensurate with the scope of protection sought by the claims. In re Moore, 439 F.2d 1232, 169 USPQ 236 (CCPA 1971). Once the scope of the claims is addressed, a determination must be made as to whether one skilled in the art is enabled to make and use the entire scope of the claimed invention without undue experimentation.

Before addressing issues raised by the Examiner, it is noted that the Examiner has made sweeping statements and conclusions without providing support therefor.

The Examiner is reminded that MPEP 2144.03 states:

> The Examiner may take official notice of facts outside of the record which are capable of instant and unquestionable demonstration as being "well-known" in the art. In re Ahlert, 424 F.2d 1088, 1091, 165 USPQ 418, 420 (CCPA 1970). . . ..

The scientific conclusions regarding what the skilled artisan knows and does not know that are set forth in the Office Action are"capable of instant and unquestionable demonstration as being "well-known" in the art. Evidence

beyond official notice by the Examiner must be provided to establish that one of ordinary skill in the art would have been led to do what applicant has done:

MPEP 2144.03 continues:

> If justified, the examiner should not be obliged to spend time to produce documentary proof. If the knowledge is of such notorious character that official notice can be taken, it is sufficient so to state. In re Malcolm, 129 F.2d 529, 54 USPQ 235 (CCPA 1942). If the applicant traverses such an assertion the examiner should cite a reference in support of his or her position.

In this instance, a reference or references supporting assertions made by the Examiner should be provided.

Analysis

The rejected claims

To focus of the remarks herein, the subject matter of the claims, particularly the independent claims is summarized:

Claim 29 is directed to methods of treatment of pathological conditions:

> A method for treating pathological conditions by treating the underlying pathology associated with inflammatory responses and secondary tissue damage associated with activation, proliferation and migration of immune effector cells by inhibiting activation, proliferation or migration of immune effector cells, comprising administering a conjugate to an animal whereby activation, proliferation, migration of the immune effector cells is inhibited wherein:
>
> the conjugate comprises a targeted agent or a portion thereof and a chemokine receptor targeting agent or a portion thereof sufficient to bind to the chemokine receptor and facilitate internalization of the conjugate;
>
> the chemokine receptor targeting agent is a chemokine, an antibody that specifically binds to a chemokine receptor or a fragment of the chemokine or antibody, wherein the chemokine, antibody or fragment thereof binds to the receptor and internalizes the targeted agent in a cell;
>
> the targeted agent or portion thereof, when internalized in a cell, alters metabolism or gene expression in the cell, regulates or alters protein synthesis in the cell, inhibits proliferation of the cell or kills the cell; and
>
> the conjugate binds to a chemokine receptor resulting in internalization of the targeted agent in cells bearing the receptor.

Claim 35 is directed to methods of targeted delivery:

A method of targeted delivery of an agent into cells that express chemokine receptors, comprising associating the agent with a chemokine receptor targeting agent, whereby the agent is internalized by the cells.

Claim 38 is directed to methods of inhibiting proliferation, migration or activation of cells bearing chemokine receptors:

A method of inhibiting proliferation, migration or activation of cells bearing chemokine receptors, comprising contacting the cells with an effective amount of a conjugate that comprises a targeted agent and a chemokine receptor targeting agent, or a portion thereof, wherein the conjugate binds to a chemokine receptor resulting in internalization of the targeted agent in cells bearing the receptor.

Claim 40 is directed to a method for treating secondary tissue damage:

A method for treating secondary tissue damage and associated disease states, comprising administering to a subject in need thereof an effective amount of a therapeutic agent that inhibits the proliferation, migration or physiological activity of secondary tissue damage-promoting inflammatory cells, wherein the therapeutic agent is a conjugate that comprises a chemokine receptor targeting agent and a targeted agent or portion thereof selected so that conjugate binds to a chemokine receptor and internalizes the targeted agent, which inhibits the proliferation, migration or physiological activity of the secondary tissue damage-promoting cells.

Claim 72 is directed to methods of inhibiting proliferation, migration or activation of immune cells:

A method for inhibiting activation, proliferation or migration of immune cells, comprising contacting immune cells with a conjugate that comprises a targeted agent or a portion thereof and a chemokine receptor targeting agent whereby activation, proliferation, migration of the immune cells is inhibited, wherein:

the targeted agent or portion thereof is a toxin;

the chemokine receptor targeting agent is a chemokine or a fragment of thereof that binds to a chemokine receptor and internalizes the targeted agent; and

the conjugate binds to a chemokine receptor resulting in internalization of the targeted agent in cells bearing the receptor.

Claim 86 is directed to methods for developing methods of treatment of inflammatory disorders:

A method of treating a disease or disorder associated with an inflammatory response, comprising:

identifying immune cells that are activated in the disease or disorder;

identifying chemokine receptors expressed on the cells;

preparing a conjugate or plurality thereof containing toxin linked to a chemokine or a plurality of chemokines that specifically bind to the identified chemokine receptors and effect or facilitate internalization of the toxin into the cells; and

contacting the immune cells with the conjugate or plurality thereof.

Background and summary of the claimed subject matter

As described in the application in great detail and summarized in the previous response, the activation, migration and proliferation of leukocytes are the hallmark of a vast number of immunomodulatory diseases. These cells are responsible for the production of inflammatory mediators and toxic molecules (such as cytokines, reactive oxygen species, metalloproteinases and cytotoxins) that are essential for the host immune defense against invading pathogens, such as bacteria and viruses. Inappropriate triggering, disregulation or over-activation of the immune response is responsible for the damage to normal host tissue witnessed in leukocyte-mediated diseases such as arthritis, multiple sclerosis, and pulmonary diseases. Leukocyte-mediated diseases also include trauma (e.g. spinal cord injury) and cancers and others. In the latter, leukocytes exert tumorigenic effects by nourishing the cancer directly or indirectly (by directing angiogenesis), by supplying chemokines and growth factors, and aiding metastasis by supplying various extracellular proteases.

Thus leukocytes are the mediators of diseases that can have combinations of allergic, autoimmune, angiogenic, inflammatory, and tumorigenic components. It must be noted that leukocytes are not necessarily the trigger of disease (which may be viral , bacterial, allergen, aberrant gene

expression, trauma etc – initiated) but the excess immune (leukocyte) response is responsible for disease manifestation and progression.

This application provides an avenue of the therapeutic intervention that exploits this common underlying response (termed an underlying pathological response in the claims). Selection of this pathway for therapeutic intervention is not new (see discussion below); what is new in this application is the mode of intervention. The instant application provides conjugates that are targeted to specific chemokine receptors. (see, *e.g.*, Arimilli *et al.* (2000) *Immunological Rev. 177*:43-51).

The instant inventors recognized that chemokines play an intimate role in these varied diseases, and, as described in the application, provide a large repertoire of molecules that interact with an array of receptors. It is the instant inventors who have identified chemokine receptors as ideal targets for delivery of therapeutics, such as toxins.

Having provided the mode of intervention, the use of chemokines as targeting agents as described herein, one of skill in the art will recognize by virtue of knowledge in the art and the disclosure in the application, that the method provides a means for treatment of any disease in which in inappropriate triggering, disregulation or over-activation of the immune response is involved.

The instant applicant is **not** claiming the concept that these diseases are linked by an underlying pathology, such concept is recognized by those of skill in the art, but is providing a new avenue of treatment that exploits the common underlying pathology.

For example, U.S. Patent No. 5,750, 565 is directed to the use of tetrahydrofurans tetrahydrothiophenes, pyrrolidines and cyclopentanes to treat inflammatory disorders by inhibiting the enzyme 5-lipoxygenase. The patent states:

> These compounds in general reduce the **chemotaxis and respiratory burst** leading to the formation of damaging oxygen radicals of polymorphonuclear **leukocytes** during an inflammatory or immune

response. The compounds exhibit this biological activity by inhibiting the enzyme 5-lipoxygenase.

And later:

Examples of immune, allergic and cardiovascular disorders include general inflammation, cardiovascular disorders including hypertension, skeletal-muscular disorders, osteoarthritis, gout, asthma, lung edema, adult respiratory distress syndrome, pain, aggregation of platelets, shock, rheumatoid arthritis, juvenile rheumatoid arthritis, psoriatic arthritis, psoriasis, autoimmune uveitis, allergic encephalomyelitis, systemic lupus erythematosus, acute necrotizing hemorrhagic encephalopathy, idiopathic thrombocytopenia, polychondritis, chronic active hepatitis, idiopathic sprue, Crohn's disease, Graves ophthalmopathy, primary biliary cirrhosis, uveitis posterior, interstitial lung fibrosis; allergic asthma; and inappropriate allergic responses to environmental stimuli such as poison ivy, pollen, insect stings and certain foods, including atopic dermatitis and contact dermatitis.

The claims recite:

a method for the treatment of inflammatory disorders" using their compounds which include cardiovascular disorders, asthma, psoriasis, adult respiratory distress syndrome, atopic dermatitis, and contact dermatitis.

U.S. Patent No. 6,140,338, a subsequent patent states that:

Chemokines are polypeptidic leukocytic migration factors having molecular weights of about 10,000, and at least 21 types of peptide families having similar structures have been found. Further, at least 7 types of the chemokine receptors to which chemokines bind exist on leukocyte, and the receptors are considered to play an important role by means of selective migration and activation of leukocyte in many inflammatory diseases [Trends in Pharmacological Sciences, 17, 209-213 (1996)]. Accordingly, substances which specifically inhibit binding of chemokines to the chemokine receptors are considered to **suppress the selective migration and activation of leukocyte** and thus be useful as pharmaceutical drugs for prevention or treatment of e.g. acute or chronic inflammatory diseases such as septicemia, pneumonia, arthritis or allergic diseases, cancer, ischemic reflow disorder, arteriosclerosis, or rejection symptoms after organ transplantation operation. Further, in recent years, the chemokine receptors have been identified to be receptors on target cells, which are important for AIDS virus (HIV) to infect to the target cells [Nature, 381, 661-666 (1996); Nature, 381, 667-673 (1996); Cell, 85, 1149-1158 (1996)].

The claims in that patent are directed to a method of treating a disease or conditions including, acute inflammatory disease, chronic inflammatory disease, chronic inflammatory disease, acquired immune deficiency syndrome, ischemic reflux disorders, and arteriosclerosis; septicemia, pneumonia, or arthritis; an allergy or rejection symptoms after an organ transplantation operation.

Subsequent to the instant application others have reached the conclusion that chemokines play a role in a diverse set of disorders by virtue of their role as causing activation, proliferation and/or migration of immune cells (see, *e.g.*, Proudfoot *et al.* (2000) *Immunology Rev. 177:*246-256; Segerer *et al.* (2000) *J Am Soc. Nephrol 11*:152-176; Armilli *et al.* (2000) *Immunolog. Rev. 177:* 43-51; Juang *et al.* (2000) *Immunolog. Rev. 177:*52-67; Gutierrez-Ramos *et al.* (2000) *Immunolog. Rev. 177:*31-42; Gerard *et al.* (2001) *Nature Immunol. 2*:108-115). These references, which are subsequent to the instant application, are provided not to establish enablement, but to demonstrate operativeness and to evidence confirmation of what is taught in the instant application and doubted by the Examiner.

In addition, subsequent publications have demonstrated that antagonizing chemokine activity (a different modality from the instantly claimed methods in which the cells involved are targeted) is effective in treating a variety of disorders that share the underlying pathology (see, *e.g.*, Ghirnikar *et al.* (2000) *J. Neuroscience Res. 59:63-73*). Subsequent references establish in recognized animal models that depletion of immune cells is an effective treatment (see, *e.g.*, Popovich *et al.* (1999) *J. Experimental Neurol. 158*:351-365; Hoover *et al.* (2000) *Immunol. 101*:501-511) for disorders, such as spinal cord injury, pulmonary immune fibrosis and others. Other references establish the role of immune cells in the pathology of a variety of disorders (discussed in detail in the application and prior response; see, also Huitinga *et al.* (1990) *J. Exp. Med. 172*:1025-1033; Hoover *et al.*

A recent publication of the subject matter of this application by the inventor that describes (as does the instant application) the role immune cells, particularly of leukocytes, in disease manifestation and progression of the seemingly unrelated disorders and diseases recited in the claims (see, McDonald *et al.* (2001) *IDrugs 4*:427-442). It is the proliferation, activation and migration of the immune cells that are targeted by the conjugates. By inhibiting proliferation, activation and/or migration thereof, a variety of disorders can be treated. In virtually all diseases, the treatment, inhibition of proliferation, activation and migration of the immune cells is the same, the difference will be the disease manifested by the treated subject.

Hence, differences among the prior art (and even subsequent art) is the choice of the choice of system with which to intervene. Some choose to intervene with inflammatory mediators (e.g., cytokines), others with angiogenic mediators (e.g., vascular endothelial growth factor). Others choose to intervene with leukocyte trafficking by exploiting the cell adhesion molecule systems (anti-selectins, anti-integrins, etc) or the chemokine system with receptor antagonists.

Still others choose to eradicate the cells involved in disease pathology (thereby eradicating the production of all noxious substances at once – and thus the apex of disease pathology) which can be achieved by targeting immune cells, a route exploited by the instant methods. The following table summarizes a variety of immune cell-depleting therapeutics for treatment of a variety of disorders:

Table 3. Examples of Cell Depleting Therapeutics

Agent	Name	Company	Indication(s)
Monoclonal Antibody	ABX-CBL[3]	Abgenix	GVHD
	Anti-CD11a[3]	Xoma	Psoriasis
Transplant Rejection			
	Campath®	Millenium/Ilex	Leukemia (CLL)
	Herceptin®	Genentech	Breast Cancer
	Nuvion[3]	PDL	GVDH
			Psoriasis

	Rituxan[1]	IDEC/Genentech	T-cell Cancers Lymphoma (NHL) Arthritis*
Ligand-Toxin Fusion Protein 00	Bexxar[2] Genimmune[3]	Coulter/Corixa Xoma	Lymphoma (NHL) Leukemia Lymphoma Autoimmune
Diseases			
	Mylotarg[1]	AHP Corp.	Leukemia
	NBI 3001[3]	Neurocrine Biosciences	Glioma
	Ontak[1]	Ligand	Lymphoma (CTCL)
	Zevalin[2]	IDEC	Lymphoma (NHL)

1: FDA approved drug; 2: Awaiting FDA approval; 3: In late stage clinical trials.
Bexxar and Zevalin incorporate a radionucleotide as the toxin moiety.
*: A recent clinical trial showed that almost complete depletion of B-cells with using combination therapy including Rituxan, was beneficial to patients with IgG RF committed rheumatoid arthritis – with no immunosupression. (Sustained Improvement in Rheumatoid Arthritis Following B-Lymphocyte Depletion., Edwards et al., (2000) 64th American College of Rheumatology Meeting. Philadelphia. Also, Rheumatology , in press).

Hence depletion of immune cells is effective for treatment of a variety of disorders.

The instant application teaches and claims the use of the chemokine system of receptors and ligands to eradicate excess numbers of ACTIVATED leukocytes subtypes. The application and DECLARATION of record establish using *in vitro* and *in vivo* animal models that the conjugates specifically target and eradicate immune cells. Having demonstrated the specificity of the targeting and the ability to internalize targeted agents, one of skill in the art would recognize that any disease involving a proliferation, migration or activation of immune cells are susceptible to treatment with the conjugates. Applicant is not claiming a cure for any disease, but is providing conjugates that demonstrably target and inhibit the cells involved in the inflammatory response. As such, the underlying pathology common to these diseases is targeted.

Rebuttal to arguments set forth by the Examiner:

1. The Examiner states:

Applicants are claiming a method of treating said pathological conditions wherein the disorder or disease state is selected from the group consisting of CNS injury, CNS inflammatory diseases,

neurodegenerative disorders, heart disease, inflammatory eye disease. inflammatory bowel diseases. inflammatory joint diseases, inflammatory kidney or renal diseases, inflammatory lung diseases, inflammatory responses associated with bacterial or viral infections and cytokine regulated cancers. Furthermore, the CNS inflammatory diseases and neurodegenerative disorders are selected from the group consisting of stroke, closed head injury, leukoencephalopathy, choriomeningitis, meningitis, adrenoleukodystrophy, AIDS dementia complex, Alzheimer's disease, Down's Syndrome, chronic fatigue, syndrome, encephalitis, encephalomyelitis and spongifonn encephalopathies.

Applicant respectfully disagrees. The discussion above and attached articles demonstrate that if one interferes with leukocyte-derived mediators, leukocyte trafficking and /or activation, then one can, by reason, treat a number of diverse diseases and conditions.

(i) A broad array of diseases are susceptible to treatment with the conjugates provided in this application.

It is urged that the specification provides no guidance of how to treat "every possible disorder associated with an inflammatory response."

As discussed above and previously, the claims are not directed to treating "every possible disorder associated with an inflammatory response." The claims are directed to methods of inhibiting activation, proliferation and migration of immune effector cells

Claim 29 is directed to methods of treatment of pathological conditions;

Claim 35 is directed to methods of targeted delivery not to methods of treatment per se;

Claim 38 is directed to methods of inhibiting proliferation, migration or activation of cells bearing chemokine receptors;

Claim 40 is directed to a method for treating secondary tissue damage, not all inflammatory disease states;

Claim 72 is directed to methods of inhibiting proliferation, migration or activation of immune cells;

Claim 86 is directed to methods for **developing** methods of treatment of inflammatory disorders.

Hence, only claim 29 and claims dependent thereon are direct to methods of treatment. Second, none of the claims are not directed to the treatment of inflammatory disorders; the claims are directed ot methods of inhibiting the proliferation, activation and/or migration of immune cells, a common underlying pathophysiological state that is known by those of skill in the art to be associated with a variety of diseases and disorders. As described in the specification in great detail, chemokines receptors are expressed on certain classes of activated cells and such cells associated with particular disorders. The methods herein exploit the dynamic nature of chemokine receptor distribution and upregulation that is the hallmark of inflammatory conditions.

As noted, 35 U.S.C. §112, first paragraph does not require "a specific example of everything *within the scope* of a broad claim." In re Anderson, 176 USPQ 331, at 333 (CCPA 1973), emphasis in original. Rather, the requirements of § 112, first paragraph "can be fulfilled by the use of illustrative examples or by broad terminology." In re Marzocci et al., 469 USPQ 367 (CCPA 1971)(emphasis added). In this instance, since the disorders and conditions share a common underlying cause, the requirements of § 112, first paragraph, have been fulfilled.

Third, known drugs for broad disease applications interfere with leukocyte functions. This **fact**, in a vast number of cases of drugs was not realized until after they were injected into patients. For example, steroids (e.g., methylprednisolone) are used in multiple conditions and do so by inhibiting reactive oxygen species and reduce the expression levels of cell adhesion molecules on various leukocytes. Steroids are administered for treatment of virtually all of the diseases in the instant claims.

Interferon treatments (used in treatment as diverse as MS and hepatitis) appear to down-regulate T –cell activation, proliferation, migration and their

production of inflammatory cytokines (clearly the effect of the cytokine which is only evident in certain micro-environments since relatively few patients respond). Non-steroid anti-inflammatory agents have broad application because they interfere with prostaglandin and reactive oxygen species synthesis from leukocytes. Unfortunately, the problem with most drugs is the **LACK** of selectivity for cellular targets --- and that's why you have side-effects.

There are a number FDA approved drugs, experimental drugs and non-therapeutic reagents that demonstrate that leukocyte-depletion *in vivo* is beneficial. For example, clodronate (dichloromethylene biphosphonate) is encapsulated in liposomes (for delivery purposes) and is engulfed by (activated) phagocytosing macrophages. The compound eradicates macrophages by inhibiting gene transcription. This compound has been used to specifically deplete macrophages in a safety clinical trial in the Netherlands showed that intraarticular administration elicited no toxic effects and clear anti-inflammatory activity in arthritis patients (Barrera, *et al.*, [2000] Arthritis Rheum 43: 1951-59). The same molecule has been shown to have beneficial effects in diverse animal disease models including EAE (the multiple sclerosis [MS] animal model, Huitinga *et* el., [1990] J Exp Med 172: 1025-33; spinal cord injury [SCI], (Popovich *et al.*, [1999] Exp Neurol 158: 35-65); pulmonary immune fibrosis (Zhang-Hoover *et al.*, [2000] Immunology 101: 501-11); emphysema (Ofulue and Ko, [1999] Am J Physiol 277(Lung Cell Mol Physiol 21): L97-L105) and uveitis (eye disease, Pouvreau *et al.*, [1998] J Neuroimmunol 86: 171-81).

The monoclonal antibody (mAb) Campath from Millenium has just been approved to treat chronic lymphocytic leukemia and is in clinical trials for B-cell lymphoma and MS. Another mAb, Nuvion from Protein Design Labs, is in clinical trails for T-cell malignancies, psoriasis and graft versus host disease (see Table 1). Both these antibodies kill T-cells (amongst others) by antibody and complement-dependent toxicity. Novantrone, a DNA intercalating agent from Immunex, which is toxic to proliferating cells including leukocytes is not only

used to treat patients with certain cancers but also with MS. However, the trouble here is the lack of specificity for target cells.

The animal model data using clodronate and the clinical use of clodronate and the FDA approved molecules (Table 1., many more in clinical development) is further testament to the fact that activated, proliferating leukocytes are directly involved (if not the architects) in the pathology of a diverse range of diseases and that their eradication is beneficial.

Table 1. Selected inflammatory and immunomodulatory diseases and conditions and therapeutic agents and models therefor

Acute Lung Injury And Acute Respiratory Distress Syndrome (ARDS)	[91, 92, 140-143]
Allergic Lung Disease	[131-133, 144, 145]
Alzheimer's Disease (AD)	[146-152]
Cancer (Growth, Angiogenesis, Metastasis)	[6, 9, 10, 116, 153-155]
Cardiovascular Disease	[156-160]
Chronic Obstructive Pulmonary Disease (COPD)	[161-164]
Graft Versus Host Disease (GVHD)	[165-168]
HIV Infection and AIDS	[24, 130, 169-171]
Inflammatory Bowel Disease (IBD)	[4, 172-174]
Inflammatory Responses To Burns, Gene Therapy, and Surgery	[175-179]
Multiple Sclerosis (MS)	[21, 22, 180, 181]
Proliferative Vitreoretinopathy	[182-184]
Psoriasis	[85, 87, 185, 186]
Rheumatoid arthritis (RA) and Osteoarthritis (OA)	[8, 187-190]
Spinal Cord Injury (SCI)	[19, 191-193]
Sepsis	[194-196]
Stroke	[197-200]
Systemic Lupus Erythematosus	[201-204]
Traumatic Brain Injury (TBI)	[205-209]
Uveitis	[210-213]

(listed references are set forth at the end of the response and also in the attached paper McDonald *et al.* (2001) *IDrug 4*:427-442).

All the above agents have therapeutic limitations that are avoided with the use of the more versatile conjugates provided herein and demonstrated and discussed in the Declaration Pursuant to 37 CFR § 1.132 of record, which is summarized below.

(ii) Leukocytes, Chemokines and Disease

Ligands and receptors of the chemokine system are the pivotal regulators of leukocyte activation, proliferation, and migration (trafficking) in health and disease. It is the persistent upregulation of this system that is inherent in a wide range of diseases (e.g., Gerard and Rollins, [2001] Nature Immunol. 2 (2); 108 –15; McDonald *et al.*, [2001] IDrugs 4: in press – see Table 1 and references therein; Proudfoot *et al.*, [2000] Immunol Rev 177: 246-56; Arimilli *et al.*, [2000] Immunol Rev 177: 43-51; Huang *et al.*, [2000] Immunol Rev 177: 52-67;Segrerer, *et al.*, [2000] J Am Soc Nephrol 11: 152-76; Gutierrez-Ramos *et al.*, [2000] Immunol Rev 177: 31-42; Gerszten *et al.*, [2000] J Lab Clin Med 136: 87-92). The infiltration of excess numbers of activated leukocytes to the site of disease is ultimately regulated by chemokines. If immune cells are a part of a process then **by definition, the chemokine system is involved.**

This refutes the statement by the Examiner that there is only "some chemokine involvement in the disorder" (claim rejection A, middle page 5) – chemokine involvement is paramount and is near the apex of the pathological processes in many diseases.

What is very apparent from the *in vitro* and *in vivo* observations described in the literature (e.g., the references cited above) is that:-

The expression of chemokines and their receptors is finely and specifically regulated at various levels (e.g., transcription and translation in different microenvironments, tissues and organs) in health and disease.

The expression of chemokine receptors is restricted to **activated** leukocytes (and a few other tissue specific cells) involved in the disease process, and in many cases are only expressed when the disease process has begun (e.g., Ghirnikar *et al*, [2000] J Neurosci Res 59: 63-73).

Specific leukocyte cell subtypes express certain chemokine receptors that are associated with specific tissues and diseases such as eosinophils and CCR3 in asthma and microglia/T-cells; CCR2/CXCR3 with MS in the central nervous system; CCR2 and macrophages in atherosclerosis; CXCR4 and CCR5 in HIV infection.

The specificity (i.e., the receptor(s) targeted) of a given chemokine-receptor targeting conjugates (**CRTCs**) provided in the instant application is dictated by its chemokine moiety. Therefore, it follows that a specific CTRC can be chosen based upon the leukocyte subtype(s) and chemokine receptors they express in a given clinical disease/condition. The experimental data summarized below indicates that CTRCs behave in a predicable fashion targeting only **activated** target cells known to express the prerequisite receptor type.

(iii) Summary of *in vitro* and *in vivo* Experimental Observations with Exemplary Chemokine-Toxins (see, DECLARATION of record)

The CRTCs designated OPL908110 and OPL98111 have been extensively characterized in tissue culture and the latter has been used in two xenograft animal model experiments. This data was provided in the previous response in the DECLARATION of record. In summary,

both exemplary conjugates target cells bearing their respective receptors (e.g., monocytes, macrophages and T-cells) and do not effect non-target cells (known not to express the prerequisite receptors).

Experiments have shown that target cells have to be in an activated state in order to be eradicated (also , see McDonald *et al.* (2001) *ID Drugs 4*:427-442).

OPL98110 targets leukocytes but **not** neurons or astrocytes in culture.

These results and the observations that MCP-1 and its cognate receptor, CCR2 are upregulated and play a direct role in MS, EAE, SCI TBI, atherosclerosis and arthritis (e.g., references cited above) indicates that OPL98110 is an ideal candidate for the treatment of these diseases.

OPL98110 eradicates breast carcinoma cells but not gliomas, whereas OPL98111 eradicates gliomas and colon cancer cells but not breast carcinoma cells.

OPL98111 eradicates glioma, colon and leukemia cells in a dose responsive manner (see IDrugs article).

OPL98111 was eradicates cancer cells and slow the rate of cancer progression in two separate xenograft colon **cancer animal model experiments**. **No toxicity** was observed upon histological examination of the liver, brain or kidneys of treated animals and no premature deaths occurred.

Also evident from the histopathology from these experiments was the fact that newly formed blood vessels (angiogenesis) was evident in non-treated tumors but was absent from treated tumors. This is consistent with the fact that receptors for OPL98111 are on activated endothelial cells forming new blood vessels and that SDF as well as other chemokines, has been shown to have direct proliferative effects on these cells, *in vitro* and *in vivo*.

Toxicology experiments were conducted with the broad acting OPL98111 (receptor widely distributed). A massive intra-peritoneal dose of 5 mg/kg had no observable effect on mice after 15 days and no discernable tissue toxicity or immunosuppression.

The same dose intravenously resulted in death and the only observable pathology was massive necrosis of the colon (high turnover of activated cells bearing the appropriate receptor, CXCR4). Liver and kidneys were unscathed. Chemokine-toxins which have a narrower receptor distribution will have even less toxicity.

OPL98111 was also shown to greatly decrease **viral load in HIV infected T-cells** (as effective as AZT and 3CT in similar assays).

Given these results, and leukocyte subtype/receptor participation in disease, OPL98111 will be a useful treatment for various conditions including cancers, HIV and arthritis.

Further,

Quiescent cells do not express receptors and therefore not targets for these compounds. This would suggest that immunosuppression will not be an issue since inactivated leukocytes are spared. Immunosuppression was not apparent in preliminary **Osprey toxicology studies** described above.

These compounds do not contain certain structural motifs (associated with certain immunotoxins) that cause vascular leak syndrome or liver toxicity.

2. The Examiner states that applicant provides:

no guidance or working examples of how to treat any or all of these diseases. Applicants only provide exemplary references which show the use of animal models which may be used to test chemokine receptor targeting conjugates. In addition, each of these diseases will require a different treatment regimen and, due to the breadth of the diseases and disorders recited in the claims, the treatment regimen would not be predictable to one of ordinary skill in the art. Furthermore, it is not understood how Applicants can treat the claimed disorders of the immune system of a patient by modulating the activation, proliferation and/or migration of inhibited immune effector cells without causing other immune-related problems to occur. Again, while Applicants describe numerous disease states in which chemokine/toxin conjugates could be used, Applicants give no guidance, or working examples for use of these compounds in treating patients who have these diseases. Applicant respectfully disagrees.

The specification provides ample guidance and examples for treating a wide variety of diseases that share this common underlying cause

The specification provides a substantial amount of guidance for selecting particular chemokines for treating a particular disease and for effecting treatment thereof. For example, Table 1 sets for a list of representative

chemokines associated with pathophysiological inflammatory responses, including secondary tissue damage, the receptor(s) they bind to, and the cell types affected by each in humans. Table 2 summarizes some exemplary chemokine-receptor targeting agents for treatment of selected diseases and conditions; table 3 provides the amino acid sequences of a variety of chemokines; table 5 provides physical properties of a variety of chemokine targeting agents; table 6 and the examples provide a dozen exemplary conjugates.

The specification also provides a detailed description of disease states associated with the inflammatory response and secondary tissue damage treatment of a provides ample guidance for the treatment of specific and classes of inflammatory disorders (see pages 151-160 and the extensive discussion in the previous response):

> Exemplary disorders and conditions, in addition to spinal cord injury, include stroke, acute lung injury and acute respiratory distress syndrome (ARDS), Alzheimer's disease, Down's syndrome, inflammatory joint disease, HIV encephalitis, growth, neovascularization (angiogenesis) and metastases of several forms of cancer including, brain, breast, and lung cancers, multiple sclerosis, spongiform encephalopathies, sepsis, ulcerative colitis and Crohn's disease, proliferative vitreoretinopathy and uveitis.

The specification describes at least five broad classes of disorders, including cancer, pulmonary diseases, viral infections, secondary tissue damage, inflammatory joint diseases and autoimmune disorders, and includes a description of at least 70 diseases that fall in one or more of these categories and describes how to select a targeting agent therefor and how to treat each diseases.

In addition, those of skill in the art, as evidenced by the large body of literature directed to chemokines, can readily identify and select an appropriate chemokine or set thereof to use based upon the teachings and guidance in the specification, which teaches how to make conjugates and exemplifies how to test them for requisite activities.

As discussed previously, the specification exemplifies quite a few conjugates, specifically demonstrates synthesis of a dozen and provides data evidencing the activity of at least two. The DECLARATION provides addition *in vitro* and *in vivo* data in recognized model systems evidencing the selectivity and specificity of the conjugates. Having demonstrated that several conjugates perform as described and predicted in the specification, there is no reason to believe that other conjugates prepared and used as taught in the specification and in accord with the claims will not have the desired properties.

The DECLARATION and Examples provide data demonstrating the activity of at least two different conjugates that rely on chemokines (MCP-1 and SDF-1β) that have very different specificity and selectivity profiles, and shows that each is efficacious for a particular type of disorder.

As noted, the specification provides lists of and tables of chemokines and exemplary fusion proteins and the diseases for which each can be used. Table 2 lists exemplary chemokine ligands and the disease to be treated. The following table lists conjugates exemplified in the specification and the disease targets and also conjugates prepared as described in the application and targeted diseases:

Table 5. Selected chemokine-toxin fusion proteins.

Chemokine	Name	Receptors	Clinical applications
Eotaxin	OPL98112	CCR1& CCR3	asthma, allergic nasal disease, IBD
MCP-1	OPL98110	CCR2	ALI*, cancers, angiogenesis, COPD, MS, SCI, T8I, uveitis
MCP-3	OPL98109	CCR1, 2 & 3	ALI*, cancers, angiogenesis, COPD, MS, SCI, T8I, uveitis
SDF-1β	OPL98111	CXCR4	arthritis, cancers, angiogenesis, HIV/AIDS
Other conjugates			
GRO-α	OPL00201	CXCR1 & 2	arthritis, COPD, cancers, angiogenesis and uveitis
IL-8	OPL00202	CXCR1 & 2	arthritis, COPD, cancers, angiogenesis and uveitis
IP-10	OPL00203	CXCR3	arthritis, GVDH, MS, SCI, Stroke,
RANTES	OPL00204	CCR1, 3, 4 & 5	arthritis, asthma, GVDH, HIV/AIDS, MS, SCI

* Acute Lung Injury

Hence the specification provides guidance in selecting the chemokine and the disease target. Furthermore, the specification describes formulation and adminstration of the conjugates (see, *e.g.*, Section F, page 142), and the DECLARATION evidences biological activity and effectiveness in *in vitro* and *in vivo* models.

In addition, examples of the method of treatment and treatment regimes that can be followed with CRTCs in a wide range of disease models are known to those of cskill in the art.

Method of Treatment and Treatment Regimes with Leukocyte-Depleting Agents

Clinical trial and animal model data on FDA approved cell-targeting agents akin to the CRTCs (mAbs and ligand-toxins) (see Table 1 above) are available from the literature and Online from Company Web Sites. Methodologies and treatment regimes are predictable to one skilled in the art.

Furthermore, the tissue culture, animal model and toxicology data evidence that the CRTCs tested thus far show similar activities in the range of like molecules discussed above. The exact dosing regimes and routes of administration of the different CRTCs depend on the indication. There is no requirement in US patent laws for clinical trials to meet the enablement standard under 35 U.S.C. §101 or §112 (see Guidelines discussed above).

Therefore, as discussed in the previous response and addressed above, it would not require undue experimentation to practice the methods as claimed.

Furthermore, it is unfair and unduly limiting to require applicant to limit the claims to specific diseases, when the application clearly describes broadly applicable methods. To do so is contrary to the public policy upon which the U.S. patent laws are based. The instant application provides methods that are generally applicable to treatment of a broad array of diseases, since the methodology is based upon targeting an underlying pathology If applicant is required to limit the claims to specific diseases, then those of skill in the art

could by virtue of the teachings of this application select chemokines and prepare conjugates for treating other diseases. To permit that is simply not fair. The instant application teaches a way of treating a whole array of disorders and conditions and, having done places the public in possession of such knowledge. Having provided this disclosure, it permits others to benefit therefrom. Those of skill in the art should not be permitted to practice what is taught in the application, but avoid infringing the claims.

3. It is alleged that it is "not predictable to one of skill in the art how to use a method of treating a patient with 'any' type of inflammatory response", because "Applicants do not give exact dosages or a treatment regimen."

As discussed above, the claim are not directed to methods for treating a patient with "any" type of inflammatory response, but to treating patients with pathophysiological responses that arise from proliferation, activation or migration of immune cells, which immune cells when proliferating or migrating or when they are activated express chemokine receptors or upregulate such receptors not present on quiescent cells. As discussed above, the specification provides ample guidance for treating such conditions.

Furthermore, the specification does provide guidance for formulation and administration of the conjugates. For example at pages 142 *et seq*, the section entitled "Formulation and administration of compositions containing the conjugates" provides substantial guidance for effecting treatment. At page 146, for example, the specification states:

> The therapeutic agents for use in the methods can be administered by any route known to those of skill in the art, such as, but are not limited to, topically, intraarticularly, intracisternally, intraocularly, intraventricularly, intrathecally, intravenously, intramuscularly, intraperitoneally, intradermally, intratracheally, as well as by any combination of any two or more thereof.
>
> The most suitable route for administration will vary depending upon the disease state to be treated, for example the location of the inflammatory condition. Modes of administration include, but are not limited to, topically, locally, intraarticularly, intracisternally, intraocularly,

> intraventricularly, intrathecally, intravenously, intramuscularly, intratracheally, intraperitoneally, intradermally, and by a combination of any two or more thereof. For example, for treatment of SCI and other CNS inflammatory conditions, local administration, including administration to the CNS fluid or into the brain (e.g., intrathecally, intraventricularly, or intracisternally) provides the advantage that the therapeutic agent can be administered in a high concentration without risk of the complications that may accompany systemic administration of a therapeutic agent. Similarly, for treatment of inflammatory joint diseases, local administration by injection of the therapeutic agent into the inflamed joint (i.e., intraarticularly) may be preferred. As another example, a disease state associated with an inflammatory skin condition may advantageously be treated by topical administration of the therapeutic agent, for example formulated as a cream, gel, or ointment. For treatment of a disease state associated with an inflammatory lung condition, the preferred route for administration of the therapeutic agent may be by inhalation in an aerosol, or intratracheally . . ..

As discussed above, the following section in the application describes at least 70 disorders that can be treated using the conjugates and in accord with the claimed methods.

4. Further, it is alleged that no guidance is provided, or working examples, for use of the claimed compounds in treating patients who have a disorder of the immune system, and it is not predictable to one of ordinary skill in the art how to treat such disorders without causing further disorders to the altered immune system.

First, it is noted that the methods herein are not designed for treating disorders of the immune system, but for treating disorders whose underlying cause is the pathophysiological inflammatory response that occurs in connection with a variety of disorders. The methods target cells of the immune system involved in other diseases not a disorder of the immune system. The specification lists more than seventy such disorders, and describes, among other spinal cord injury, which is representative and exemplary, in detail. As stated on described on pages 152 *et seq.*:

> It has been found herein that the cell biology of more than seventy diseases and conditions, involving most organ systems, involved

pathophysiological inflammatory responses in a manner similar to the cell biology of acute SCI. The following, non-exhaustive list, and the more detailed treatment of four clinical areas, are designed to illustrate some of the more important similarities. Exemplary disorders and conditions, in addition to spinal cord injury, include stroke, acute lung injury and acute respiratory distress syndrome (ARDS), Alzheimer's disease, Down's syndrome, inflammatory joint disease, HIV encephalitis, growth, neovascularization (angiogenesis) and metastases of several forms of cancer including, brain, breast, and lung cancers, multiple sclerosis, spongiform encephalopathies, sepsis, ulcerative colitis and Crohn's disease, proliferative vitreoretinopathy and uveitis.

As described in the specification and in the DECLARATION, chemokine receptors are expressed on cells, such as various leukocyte subtypes, that participate in such responses. Hence, the eradication or inhibition of such pathophysiologically upregulated cells should not cause further damage to the immune system. As described, the chemokines and chemokine receptors constitute a large family, so that the chemokine can be selected based upon the cell and particular receptor specifically expressed on the cell. The specification provides exemplary lists of chemokines and identifies the cells upon which they are regulated and the disorders for which the chemokines could be used as targeting agents.

As described in the specification, and supported by the data in the DECLARATION, targeting delivery of toxins to chemokine-bearing cells provides a means for specific targeted delivery of toxins. The *in vitro* data and *in vivo* xenograft mouse model data shows that the conjugates are specifically targeted to activated cells and do not interact with quiescent cells.

Furthermore, the *in vivo* data presented evidences the relatively low toxicity of the conjugates. The data provided in the DECLARATION shows that even high doses of OPL98111 do not completely eradicate primary human monocytes in culture, since not all of them are in the activated state. Also, a massive (non-therapeutic dose) IP dose (5 mg/kg) of OPL98111 had no apparent effect on the health of mice that were not sacrificed until over 3 weeks after treatment. Throughout the forty days of the course of experiment, mice

receiving multiple doses of OPL98111 in two xenograft experiments exhibited no difference in health when compared to placebo treated mice.

B. Claims 29, 31, 32, 34, 38, 40, 42, 44, 46, 48-54, 57 and 65-85 are rejected for the reasons already of record as containing subject matter which is not described in the specification. The Examiner urges:

> Applicant provides no guidance or working examples of how a portion of a targeting agent and/or a targeted agent can effectively bind to a cell bearing the necessary receptor and internalize, or how a portion of a targeted agent can treat a disorder associated with inflammatory responses associated with an immune effector cell. Furthermore, it is also unpredictable to one of ordinary skill in the art as to what portions of said targeting agents have the necessary activity to bind chemokine receptor to cause internalization of the targeting agent/targeted agent conjugate, or what portions of the targeted agents have the necessary activity to produce toxic effects in all cells involved in all disorders. It has been shown that the function of a peptide cannot be determined based solely on knowing the amino acid sequence (see Rudinger *et al.* 1976, especially the conclusion). The possible effect of changing even one amino acid in a polypeptide can be seen in Cunningham and Wells (1989; Abstract) in which certain single substitutions of alanine in various positions of human growth hormone dramatically altered its binding affinity for the human growth hormone receptor. In addition. George et al. (1988; p. 145) states that: "methods will not be able to assess biological relatedness until the structure/function problem is more clearly understood."

The Examiner concludes that the breadth of the claims is too extensive regarding all portions of all targeting gents. In addition, there is a lack of guidance or working examples of how a portion, or a sufficient portion of a targeting agent and/or a targeted agent can effectively bind to a cell bearing the necessary receptor and internalize, or how a portion of a targeted agent can treat a disorder associated with inflammatory responses associated with an immune effector cell. Furthermore, it is also unpredictable to one of skill in the art as to what portions of said targeting agents have the necessary activity to bind chemokine receptor to cause internalization of the targeting agent/targeted agent conjugate, or what portions of the targeted agents have the necessary

activity to produce toxic effects in all cells involved in all disorders. For these reasons, the Examiner maintains that undue experimentation would be necessary to practice the claimed methods. This rejection is respectfully traversed.

Rebuttal

First it is noted that the Examiner's arguments directed to the effect of replacing a single amino acid or a plurality thereof on protein function is not apt in the instant case. The instant claims recite that the portion of the chemokine is one that permits binding the receptor and internalization. Such portion can be readily determined by systematically truncating the proteins from one end or another. Applicant is not claiming the chemokines. These are well-known proteins whose functions, structures and activities are known. If not known, can be readily ascertained by routine experimentation. The references relied upon by the Examiner to support his inapt premise are a 1976, 1988 and 1989 references. Such references do not establish the state of the art at the time of effective filing date of the instant application in **1998**. There has been a tremendous increase in knowledge since 1976, and the 1980s.

As stated in the previous response, the specification defines what is meant by a "portion" and the functionally define the term for the chemokine receptor targeting agent and for the targeted agent:

> As used herein a portion of a chemokine refers to a fragment or piece of chemokine that is sufficient, either alone or as a dimer with another fragment or a chemokine monomer, to bind to a receptor to which chemokine dimers bind and internalize a linked targeted agent.
>
> As used herein, an amino acid residue of chemokine is non-essential if a chemokine dimer in which one or both chemokine monomers have been modified by deletion of the residue possesses substantially the same ability to bind to a chemokine receptor and internalize a linked agent that the dimer has with the amino acid(s).

In the previous response it was noted that the claims do not merely recite "a portion", but include functional language that states that the portion is sufficient to bind to a targeted receptor and facilitate internalization. The standard for undue breadth of claims is whether it would require undue experimentation to practice what is claimed. It would not require undue experimentation to select a portion effective to do what is claimed. Furthermore, the data provided in the DECLARATION was obtained using conjugates that contain only "a portion" of a toxin subunit.

With respect to the receptor targeting agent, the claims recite that portion is for specific binding and internalization of linked targeted agents. It would not require undue experimentation to identify portions of a chemokine or chemokine receptor-targeting agent that would achieve the desired result.

One of skill in the art in 1998, with the advances in bioinformatics and the ability to model protein structures, one skilled in the art could assess and identify modifications that alter binding capacities, especially since sites necessary for receptor interaction are published and known to those of skill in the art. Furthermore, from a practical standpoint, only active portions of either moiety will be selected for drug manufacture. Truncation and modification of the targeting agent or targeted agent are routinely performed in this art.

For truncated targeting agents see, *e.g.*:

A modified (circularly permuted) form of IL-4 is used in chimeras that have increased activity over the wild type and is also fused to a truncated form of PE toxin (Krietman*et al.* (1994) *Proc. Natl. Acad. Sci. U.S.A. 91*:6889-93; Puri*et al.* (1996) *Cancer Res* 56:5631-37 (both moieties truncated);

A biologically active chimera has been successfully synthesized by using a mutant FGF-2 with Cys96 replaced by Se ([S96]FGF-2) and a recombinant saporin mutant containing a single Cys at the -1 position (Buechler *et al.* (1995) *Eur. J. Biochem* 234:706-13 both moieties);

Several biologically active forms of chemokines with single amino acid substitutions have been reported including RANTES (International PCT application No.WO 98/13495), MIP-1α and MIP-1β (Hunter *et al.* (1995) *Blood* 86:4400-8; Czaplewski, *et al.* (1999) *J. Biol. Chem. 274*:16077-84). There is no reason to believe that there receptor binding capacities would be compromised if a toxin were conjugated to the chemokine C-terminus. N-and C-terminal truncated chemokines (natural or artifact) have been reported that have favorable alterations in their biological activity (*e.g.*, Weber *et al.* (1996) *J. Exp. Med.* 183:681-5; Struyf,*et al.* (1998) *J. Immunol.* 161:2672-5; Wuyts, *et al.* (1999) *Eur. J. Biochem 260*: 421-9; Wuyts, *et al.* (1999) *J. Immunol.* 163:6155-63). Therefore, contrary the assertion of the Examiner, it would be routine to make truncated targeted and targeting agents that result in conjugates that bind to targeted receptors and internalize the linked targeting agents.

Furthermore, the instant application exemplifies and provides data using a truncated shiga toxin and teaches (see, *e.g.*, page 82) that truncated toxins have been used "Truncated forms and mutants of the toxin enzymatic subunits can also be used as a cell toxin moiety (Pastan *et al.*, *Annu. Rev. Biochem. 61*:331-54; Brinkmann and Pastan, *Biochim. et Biophys. Acta 1198*:27-45, 1994; Mesri *et al., J. Biol. Chem. 268*:4852-62, 1993; Skinner *et al., Microb. Pathog. 24*:117-22, 1998; and U.S. Patent No. 5,082,927)".

For other truncated targeted agents see, *e.g.*:

Volk *et al.* cited by the Examiner; Volk *et al.* only uses a truncated PE toxin);

the exemplified targeted agents include truncated Shiga toxin that is a portion of a portion of a parent compound, exemplified conjugates contain residues 23-268 of the shiga A1 subunit;

Ontak® from Ligand Pharmaceuticals contains a portion/fragment of Diphtheria toxin (Met1-Thr387) (see, also Williams *et al.* (1990) *J Biol Chem* 265:11885-11889).

As discussed previously, the factors to be considered include, the quantify of experimentation required, the level of skill in the art, the knowledge of those of skill in the art, the guidance in the specification, presence of working examples predictability and breadth of the claims. In this instance, there is a substantial amount of guidance provided in the specification, which teaches and exemplifies assays, cytotoxicity and cell based binding assays, to use to test the conjugates and criteria to use for selecting them. One of skill in the art could readily systematically delete portions of a selected chemokine to identify the minimal or requisite portion of a chemokine or other such agent that would effectively bind to a receptor on a cell by assessing binding and effect internalization by assessing cytotoxicity or looking for internalization of a linked label. As previously noted, the level of skill in this art is recognized to be high and the knowledge of those of skill in the art is extensive, as evidenced by the body of literature, much of it cited in the specification, regarding the identifies, specificities, properties and sequences of chemokines and other chemokine-receptor targeting agents.

Thus, it would be unfair and unduly limiting to require applicant to exclude from the claims portions of chemokine-receptor targeting agents that permit binding and facilitate internalization, when those of skill in the art could readily do so. If the claims are so limited, those of skill in the art could avoid infringement merely by making routine modifications of the chemokine targeting agents, thereby using the instant disclosure but avoiding infringement.

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* * *

In view of the above amendments and remarks, reconsideration and allowance of the application are respectfully requested.

Respectfully submitted,
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IN THE UNITED STATES PATENT AND TRADEMARK OFFICE

Applicant: McDONALD et al.

Serial No.: 09/360,242

Filed: July 22, 1999

For: *METHODS AND COMPOSITIONS FOR TREATING SECONDARY TISSUE DAMAGE AND OTHER INFLAMMATORY CONDITIONS AND DISORDERS*

Art Unit: 1646

Examiner: Landsman, R.

I hereby certify that this paper and the attached papers are being deposited with the United States Postal Service as first class mail in an envelope addressed to: Commissioner for Patents Washington, D.C. 20231, on this date.

06/20/01
Date

Stephanie Seidman

ATTACHMENTS TO THE RESPONSE

1. Marked up claims (37 C.F.R. § 1.121)

2. Copies of the stamped received Postcard, Supplemental Response, and executed DECLARATION, mailed on September 11, 2000.

3. Copies of references discussed in the response

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06 20 01
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Stephanie Seidman

MARKED UP CLAIMS (37 C.F.R. § 1.121)

Please amend claims 29, 31, 65, 72 and 86 as follows:

29. (Twice Amended) A method for treating pathological conditions by treating the underlying pathology associated with inflammatory responses and secondary tissue damage associated with activation, proliferation and migration of immune effector cells by inhibiting activation, proliferation or migration of immune effector cells, comprising administering a conjugate to an animal whereby activation, proliferation, migration of the immune effector cells is inhibited[, thereby ameliorating an underlying pathology associated with adverse inflammatory responses and secondary tissue damage], wherein:

the conjugate comprises a targeted agent or a portion thereof and a chemokine receptor targeting agent or a portion thereof sufficient to bind to the chemokine receptor and facilitate internalization of the conjugate;

the chemokine receptor targeting agent is a chemokine, an antibody that specifically binds to a chemokine receptor or a fragment of the chemokine or antibody, wherein the chemokine, antibody or fragment thereof binds to the receptor and internalizes the targeted agent in a cell;

the targeted agent or portion thereof, when internalized in a cell, alters metabolism or gene expression in the cell, regulates or alters protein synthesis in the cell, inhibits proliferation of the cell or kills the cell; and

the conjugate binds to a chemokine receptor resulting in internalization of the targeted agent in cells bearing the receptor.

31. (Amended) The method of claims 29, wherein the treated pathology underlies a disorder or disease state that is selected from the group consisting of CNS injury, CNS inflammatory diseases, neurodegenerative disorders, heart disease, inflammatory eye diseases, inflammatory bowel diseases, inflammatory joint diseases, inflammatory kidney or renal diseases, inflammatory lung diseases, inflammatory nasal diseases, inflammatory thyroid diseases, inflammatory responses associated with bacterial or viral infections and cytokine-regulated cancers.

65.(Amended) The method of claim 29, wherein the chemokine receptor targeting agent is a chemokine or a sufficient portion thereof to specifically bind to a chemokine receptor and to facilitate internalization of the conjugate.

72. (Amended) A method for inhibiting activation, proliferation or migration of immune cells, comprising contacting immune cells with a conjugate that comprises a targeted agent or a portion thereof and a chemokine receptor targeting agent[or portion thereof], whereby activation, proliferation, migration [or] of the immune cells is inhibited, wherein:

the targeted agent or portion thereof is a toxin;

the chemokine receptor targeting agent is a chemokine or a fragment of thereof that binds to a chemokine receptor and internalizes the targeted agent; and

the conjugate binds to a chemokine receptor resulting in internalization of the targeted agent in cells bearing the receptor.

86. (Amended) A method of treating a disease or disorder associated with an inflammatory response, comprising:

identifying immune cells that are activated in the disease or disorder;

identifying chemokine receptors expressed on the cells;

preparing a conjugate or plurality thereof containing toxin linked to a chemokine or a plurality of chemokines that specifically bind to the identified chemokine receptors and effect or facilitate internalization of the toxin into the cells; and

contacting the immune cells with the conjugate or plurality thereof.